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Karen J. Fildes

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School of Biological Sciences

Pesticide exposure in free-living native birds and the effects of acute dosing of fenitrothion and fipronil on physiological performance in selected species

Karen Fildes BABSc (Hons)

**"This thesis is presented as part of the requirements for the
Award of the Degree of Doctor of Philosophy
of the
University of Wollongong"**

September 2008

CERTIFICATION

I, Karen Josephine Fildes, declare that this thesis, submitted in fulfilment of the requirements for the award of Doctor of Philosophy, in the School of Biological Sciences, University of Wollongong, is wholly my own work unless otherwise referenced or acknowledged. The document has not been submitted for qualifications at any other academic institution.

ABSTRACT

Chapter 1

This chapter introduces the main classes of pesticides with a brief overview of their mode of toxic action. Cholinesterase inhibiting chemicals and their known effects on birds are reviewed in detail. The benefits and limitations of the use of lethal compared to sublethal toxicological endpoints to assess pesticide impacts on avian species in toxicological research are discussed. It is argued that there is a critical need for investigations of sublethal effects to consider biochemical and physiological components of ecologically relevant traits. The use of pesticides for locust control, particularly fenitrothion and fipronil, by the Australian Plague Locust Commission, is discussed. The chemical properties and mode of toxic action of these two pesticides are reviewed and the potential impact of their application on Australian native bird species is assessed.

Chapter 2

Cholinesterase (ChE) inhibiting pesticides are applied throughout Australia to control agricultural pests. Blood plasma ChE activity is a sensitive indicator of exposure to organophosphorus insecticides in vertebrates. To aid biomonitoring and provide reference data for wildlife pesticide-risk assessment, plasma acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) activities were characterised in nine native bird species: brown songlarks (*Cinclorhamphus cruralis*), budgerigars (*Melopsittacus undulatus*), clamorous reed warblers (*Acrocephalus stentoreus*), double-barred finches (*Taeniopygia bichenovii*), king quails (*Coturnix chinensis*), Richard's pipits (*Anthus novaeseelandiae*), white-plumed honeyeaters (*Lichenostomas penicillatus*), willie wagtails (*Rhipidura leucophrys*) and yellow

throated miners (*Manorina flavigula*). The plasma of all species contained AChE and BChE except in king quail where no AChE was present. The lowest detectable plasma AChE activity was 0.10 $\mu\text{mol}/\text{min}/\text{ml}$ in budgerigars and the highest was 0.86 $\mu\text{mol}/\text{min}/\text{ml}$ in clamorous reed-warblers. BChE activity in the plasma ranged from 0.37 in double-barred finches to 0.90 $\mu\text{mol}/\text{min}/\text{ml}$ in white-plumed honeyeaters and clamorous reed-warblers. The lowest proportion of AChE was found in budgerigars (12.8%) and highest in willie-wagtails (67.8%). Apart from king quail AChE activities in all species were within the range reported for other avian species. The absence of AChE in king quail has not previously been reported for any bird species.

The effect of sampling time on plasma ChE was assessed in budgerigars and zebra finches (*Taeniopygia guttata*) and seasonal effects were examined in zebra finches. No diurnal variation in ChE activity was found at any time of day in either species although there was a significant difference in all ChE activity between seasons in zebra finches.

Chapter 3

Huge aggregations of flightless locust nymphs pose a serious threat to agriculture when they reach plague proportions, but provide a very visible and nutritious resource for native birds. Locust outbreaks occur in spring and summer months in semiarid regions of Australia. Fenitrothion, an organophosphate pesticide, is aerially sprayed to control locust plagues. To evaluate fenitrothion exposure in birds attending locust outbreaks, we measured total plasma cholinesterase (ChE), butyrylcholinesterase (BChE) and acetylcholinesterase (AChE) activities in four

avian species captured pre- and post-fenitrothion application and ChE reactivation in birds caught post spray only. Eleven of 21 plasma samples from four species had ChE activity below the diagnostic threshold (two standard deviations below the mean ChE activity of pre-spray samples). Granivorous zebra finches (*Taeniopygia guttata*) and insectivorous white-winged trillers (*Lalage sueurii*) had significantly lower mean plasma total ChE, BChE, and AChE activity post-spray, while two other insectivores, white-browed (*Artamus superciliosus*) and masked woodswallows (*Artamus personatus*), did not. Cholinesterase was reactivated in 19 of the 73 plasma samples collected, and in one of three brain samples. We conclude that native bird species are exposed to fenitrothion during locust control operations. This exposure could have detrimental impacts as locust outbreaks and avian reproductive events are both stimulated by heavy summer rainfall, leading to co-occurrence of locust-control and avian breeding activities.

Chapter 4

The effect of fenitrothion exposure on birds was examined by measuring aerobic metabolism, blood haemoglobin (Hb) content, plasma cholinesterase activities and body mass for up to 21 d post-dose. Peak metabolic rate (PMR) was measured in a flight metabolic chamber in three dose groups of house sparrows (*Passer domesticus*) (100 mg/kg = high, 60 mg/kg = medium, 30 mg/kg = low), and one dose group each of zebra finches (*Taeniopygia guttata*) (3 mg/kg) and king quails (*Coturnix chinensis*) (26 mg/kg). Aerobic metabolism was measured during a 1 h exposure to sub-freezing thermal conditions in low dose house sparrows and king quails (26 mg/kg). Fenitrothion had no effect on metabolic rate during cold exposure or on Hb at any time. By contrast, aerobic performance during exercise in sparrows was

reduced by 58% (high), by 18% (medium), and by 20% (low) 2 d post dose. House sparrows (high) had the longest recovery period for PMR (21 d) and plasma cholinesterase (ChE) activity (14 d). House sparrows (high) and treated king quails had significantly lower body weight at 48 h post-dose whereas body mass was invariant in zebra finches and house sparrows (medium and low). Cholinesterase was maximally inhibited at 6 h post dose, and had recovered within 24 h, in house sparrows (low), king quails and zebra finches. Exercise PMR in zebra finches and king quails was reduced by 23% at 2 and 3 d post-dose, respectively, despite these birds being asymptomatic in both behaviour and plasma ChE activities.

Chapter 5

We examined the sublethal effects of the fipronil based pesticide Adonis 3UL ® insecticide on birds by measuring exercise induced peak metabolic rate (PMR) in zebra finch (*Taeniopygia guttata*) and king quail (*Coturnix chinensis*), and during a 1-h exposure to sub-freezing conditions in king quail. Exercise induced peak metabolic rate was measured in zebra finch pre-dose and at one, two, ten and twenty days after treated birds ingested 17.5 mg/kg Adonis 3UL ® mixed with canola oil and control birds received canola oil alone. Peak metabolic rate measurements were taken during exercise pre-dose and two, six and fourteen days after king quail received 30 mg/kg Adonis 3UL ® or canola oil alone. Peak metabolic rate after was not affected by fipronil in Adonis 3UL ® or by sham treatment in birds of both species. We conclude that the administered sublethal dose of fipronil did not affect exercise performance in zebra finch or in king quail nor was there evidence of fipronil induced thermoregulatory effects in king quails.

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